

Applicant : Leif Andersson et al.  
Serial No. :  
Filed :  
Page : 6

Attorney's Docket No.: 11145-023US1

REMARKS

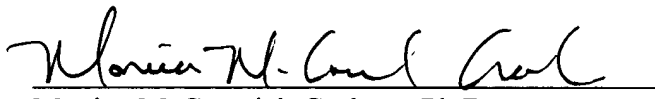
Applicants have canceled claims 34-37 without prejudice to continued prosecution.  
Claims 4-5, 9-11, 13-26, 28-29, 31, and 33 have been amended to remove multiple dependencies.  
No new matter has been added.

Attached is a marked-up version of the changes being made by the current amendment.

Applicant asks that claims 1-33 be examined. Enclosed is a \$562.00 check for the filing and excess claim fees. Please apply any other charges or credits to Deposit Account No. 06-1050.

Respectfully submitted,

Date: 3/8/02

  
Monica McCormick Graham, Ph.D.  
Reg. No. 42,600

Fish & Richardson P.C., P.A.  
60 South Sixth Street  
Suite 3300  
Minneapolis, MN 55402  
Telephone: (612) 335-5070  
Facsimile: (612) 288-9696

**Version with markings to show changes made**

**In the claims:**

Claims 34-37 have been canceled.

Claims 4-5, 9-11, 13-26, 28-29, 31 and 33 have been amended as follows:

4) (Amended) A polypeptide of [any of] claim[s] 1 [to 3], wherein said polypeptide comprises the sequence of SEQ ID NO: 2 or SEQ ID NO:4.

5) (Amended) A polypeptide of claim [4] 1, wherein said polypeptide comprises the sequence of SEQ ID NO: 28, SEQ ID NO: 30 or SEQ ID NO: 32.

9) (Amended) A polypeptide of claim 8 selected [among:  
-] from the group consisting of a polypeptide having a sequence resulting from a R→Q substitution at a position corresponding to position 41 [in] of SEQ ID NO: 2;  
[-] and a polypeptide having a sequence resulting from a V→I substitution at the position corresponding to position 40 of SEQ ID NO: 2.

10) (Amended) A polypeptide which is a mutant of a gamma subunit of a vertebrate AMP-activated kinase, wherein said polypeptide results from a deletion of a part of a polypeptide of [any of] claim[s] 1 [to 5].

11) (Amended) A nucleic acid sequence encoding a polypeptide of [any of] claim[s] 1 [to 10], or the complement thereof, provided that said nucleic acid sequence does not consist of the EST GENBANK AA178898, or of the EST W94830.

13) (Amended) A nucleic acid sequence comprising at least a portion of a nucleic acid sequence encoding a polypeptide of [any of] claim[s] 1 [to 10], and up to 500 kb of a 3' and/or of a 5' adjacent genomic DNA sequence, or the complement thereof.

14) (Amended) A nucleic acid fragment selected [among:

- a specific fragment of a nucleic acid sequence encoding a polypeptide of any of claims 1 to 10] from the group consisting of:

a) a specific fragment of a nucleic acid sequence encoding a gamma subunit of a vertebrate AMPK, wherein said gamma subunit is a polypeptide comprising at least a sequence having at least 70% identity with the polypeptide SEQ ID NO: 2,

b) a specific fragment of a nucleic acid sequence encoding a polypeptide which is a functionally altered mutant of a gamma subunit of a vertebrate AMPK, wherein said polypeptide has at least a mutation located within the first CBS domain of said gamma subunit,

c) a specific fragment of a nucleic acid sequence encoding a polypeptide which is a mutant of a gamma subunit of a vertebrate AMPK, wherein said polypeptide results from a deletion of a part of a polypeptide of a) or b);

[or of] d) a nucleic acid sequence [of claim 13] comprising at least a portion of a nucleic acid sequence encoding a polypeptide of a), b), or c), and up to 500 kb of a 3' and/or of a 5' adjacent genomic DNA sequence, or the complement thereof; and

[-] e) a nucleic acid fragment which specifically [hybridises] hybridizes under stringent conditions with a nucleic acid sequence [encoding a polypeptide of any of claims 1 to 8, or of a nucleic acid sequence of claim 11] of a), b), c), or d);

provided that said nucleic acid fragment does not consist of the EST GENBANK AA178898 or of the EST GENBANK W94830.

15) (Amended) A set of primers for amplifying a nucleic acid sequence [of any of claims 11 to 13 or a portion thereof], comprising [at least] a primer consisting of a nucleic acid fragment of claim 14.

16) (Amended) A recombinant vector comprising a nucleic acid sequence encoding a polypeptide of [any of] claim[s] 1 [to 10].

17) (Amended) [An] A host cell transformed by a nucleic acid sequence encoding a polypeptide of [any of] claim[s] 1 [to 10].

18) (Amended) A transgenic animal transformed by a nucleic acid sequence encoding a polypeptide of [any of] claim[s] 1 [to 10].

19) (Amended) A knockout animal, wherein the gene encoding a polypeptide of [any of] claim[s] 1 [to 5] is inactive.

20) (Amended) A heterotrimeric AMPK wherein the  $\gamma$  subunit consists of a polypeptide of [any of] claim[s] 1 [to 10].

21) (Amended) A method of detecting a metabolic disorder resulting from a mutation in a gene encoding a  $\gamma$  subunit of AMPK, wherein said process comprises:

[-] a) obtaining a nucleic acid sample from a vertebrate;

[-] and

b) checking the presence in said nucleic acid of a nucleic acid sequence encoding a polypeptide of [any of] claim[s] 1 [to 10], wherein said polypeptide is functionally altered.

22) (Amended) A method of claim 21 wherein the disorder is correlated with an altered glycogen accumulation in the muscular cells and results from the expression of a functionally altered allele of a polypeptide [of any of claims 1 to 5] , wherein said polypeptide comprises a sequence of SEQ ID NO:2, SEQ ID NO:4, SEQ ID NO:28, SEQ ID NO:30, or SEQ ID NO:32.

23) (Amended) A method of [any of] claim[s] 21 or] 22 wherein the presence of the nucleic acid sequence encoding said mutant polypeptide is checked by contacting said nucleic acid sample with a nucleic acid probe [obtained from a nucleic acid of claim 14 and] spanning said mutation, under conditions of specific [hybridisation] hybridization between said probe and the mutant sequence to be detected, and detecting the [hybridisation] hybridization complex.

24) (Amended) A method for obtaining a pair of primers allowing to detect a genetic polymorphic marker linked to a nucleic acid sequence encoding a polypeptide of [any of] claim[s] 1 [to 5], wherein said process comprises:

[-] a) screening a genomic DNA library from a vertebrate with a probe specific for a nucleic acid sequence encoding [a] said polypeptide [of any of claims 1 to 5,] in order to select clones comprising said nucleic acid sequence and flanking chromosomal sequences;

[-] b) identifying a polymorphic locus in said flanking chromosomal sequences, and sequencing a DNA segment comprising said polymorphic locus;

[-] and

c) designing primer pairs flanking said polymorphic locus.

25) (Amended) A method of claim 24 wherein the selected clones comprise at least a portion of a nucleic acid sequence encoding [a] said polypeptide [of any of claims 1 to 5] and up to 500 kb of a 3' and/or of a 5' adjacent sequence.

26) (Amended) A method of [any of] claim[s] 21 [to 25] wherein the vertebrate is a mammal.

28) (Amended) A pair of primers obtainable by the process of [any of] claim[s] 24 [to 26].

29) (Amended) A process for detecting a dysfunction of carbohydrate metabolism resulting from the expression of a functionally altered allele of a polypeptide of [any of] claim[s] 1 [to 5] in a vertebrate, wherein said process comprises:

[-] a) obtaining a sample of genomic DNA from said vertebrate;

[-] b) contacting said DNA with a pair of primers [of claim 28] under conditions allowing PCR amplification;

[- analysing] and

c) analyzing the PCR product to detect if an allele of a polymorphic marker linked to a nucleic acid sequence encoding a functionally altered allele of [a] said polypeptide [of any of claims 1 to 5] is present.

31) (Amended) A process of [any of] claim[s 29 or] 30, wherein said vertebrate is a mammal.

33) (Amended) A process of claim 32 wherein the pair of primers is selected [among:  
-] from the group consisting of

a pair of primers consisting of SEQ ID NO: 5 and SEQ ID NO: 6;

[-] a pair of primers consisting of SEQ ID NO: 7 and SEQ ID NO: 8;

[-] a pair of primers consisting of SEQ ID NO: 9 and SEQ ID NO: 10;

[-] a pair of primers consisting of SEQ ID NO: 11 and SEQ ID NO: 12;

[-] a pair of primers consisting of SEQ ID NO: 13 and SEQ ID NO: 14;

[-] a pair of primers consisting of SEQ ID NO: 15 and SEQ ID NO: 16;

[-] a pair of primers consisting of SEQ ID NO: 17 and SEQ ID NO: 18;

[-] a pair of primers consisting of SEQ ID NO: 19 and SEQ ID NO: 20;

[-] a pair of primers consisting of SEQ ID NO: 21 and SEQ ID NO: 22;

[-] a pair of primers consisting of SEQ ID NO: 23 and SEQ ID NO: 24;

[-] and

a pair of primers consisting of SEQ ID NO: 25 and SEQ ID NO: 26.